```
=> d his

     (FILE 'HOME' ENTERED AT 10:48:25 ON 21 JUN 2002)

     FILE 'MEDLINE' ENTERED AT 10:48:31 ON 21 JUN 2002
L1         15333 S PKC?
L2           749 S PKCEPSILON OR PKC-EPSILON
L3         64186 S ANXIETY
L4             8 S L1 (S) L3
L5         15333 S L1 OR L2
L6           551 S PKC EPSILON
L7             0 S L6 AND L3
L8             0 S L6 (S) L3
L9             8 S L1 AND L4
L10        32580 S TRANSGENIC
L11           98 S L1 (S) L10
L12        15442 S TRANSGENIC MOUSE
L13           68 S L1 (S) L12
L14            6 S L6 (S) L12

     FILE 'MEDLINE, CAPLUS, BIOSIS, EMBASE, USPATFULL, PCTFULL' ENTERED AT
     11:00:36 ON 21 JUN 2002
L15            4 S L7
L16            4 S L8
L17            3 DUP REM L15 L16 (5 DUPLICATES REMOVED)

=> s l14
L18           31 L14

=> s dup rem l15 l16 l18
MISSING OPERATOR REM L15
The search profile that was entered contains terms or
nested terms that are not separated by a logical operator.

=> dup rem l15 l16 l18
PROCESSING COMPLETED FOR L15
PROCESSING COMPLETED FOR L16
PROCESSING COMPLETED FOR L18
L19           18 DUP REM L15 L16 L18 (21 DUPLICATES REMOVED)
```

HL-28958 (NHLBI)
HL-49537
SOURCE: CIRCULATION RESEARCH, (1996 Apr) 78 (4) 724-36.
Journal code: 0047103. ISSN: 0009-7330.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199607
ENTRY DATE: Entered STN: 19960719
Last Updated on STN: 20000303
Entered Medline: 19960711

AB The consequences of endothelin receptor activation were examined in atrial tumor myocytes derived from **transgenic mice** (AT-1 cells). Endothelin-1 (endothelin) stimulates phosphoinositide hydrolysis in a dose-dependent manner. Endothelin also induces the rapid and transient translocation of protein kinase C (**PKC**)-**epsilon** immunoreactivity from the soluble to the particulate cell fraction. The subcellular distributions of PKCalpha and PKCzeta (also expressed by AT-1 cells) are not influenced by endothelin. Using quantitative fluorescence microscopy with fura 2, we examined the effects of endothelin on intracellular calcium. In electrically driven myocytes, endothelin induces a rapid and transient increase in the amplitude of the calcium transient. This is blocked by both phorbol 12-myristate 13-acetate (PMA) pretreatment to downregulate PKC and the PKC inhibitor chelerythrine, arguing that PKCepsilon plays a critical role in endothelin receptor-dependent increases in intracellular calcium. Endothelin also stimulates mitogen-activated protein kinase (MAPK). MAPK activation is markedly attenuated by pretreatment with PMA or pertussis toxin (PTX, to activate susceptible G protein alpha subunits); it is completely prevented by combined pretreatment with PMA and PTX. In contrast, it is not attenuated by chelation of intracellular calcium with BAPTA. These findings indicate that the pathway for endothelin receptor stimulation of MAPK involves PKCepsilon and PTX-sensitive G protein(s). Thus, these studies identify a functional role for PKCepsilon as a mediator of endothelin receptor-dependent increases in cytosolic calcium and MAPK activity in AT-1 cells. Accordingly, the AT-1 cell system should provide a uniquely useful model to identify the intracellular targets for PKCepsilon and investigate their function in the regulation of intracellular calcium homeostasis and the induction of the growth response in cardiac myocytes.

L19 ANSWER 1 OF 18 USPATFULL DUPLICATE 1

ACCESSION NUMBER: 2002:88458 USPATFULL
TITLE: Use of inhibitors of protein kinase C epsilon to treat pain
INVENTOR(S): Messing, Robert O., Foster City, CA, United States
Levine, Jon D., San Francisco, CA, United States
PATENT ASSIGNEE(S): The Regents of the University of California, Oakland, CA, United States (U.S. corporation)

	NUMBER	KIND	DATE
PATENT INFORMATION:	US 6376467	B1	20020423
APPLICATION INFO.:	US 1999-347370		19990706 (9)

	NUMBER	DATE
PRIORITY INFORMATION:	US 1998-103763P	19981009 (60)
	US 1998-97755P	19980706 (60)

DOCUMENT TYPE: Utility
FILE SEGMENT: GRANTED
PRIMARY EXAMINER: Russel, Jeffrey E.
LEGAL REPRESENTATIVE: Cooley Godward LLP
NUMBER OF CLAIMS: 4
EXEMPLARY CLAIM: 1
NUMBER OF DRAWINGS: 29 Drawing Figure(s); 11 Drawing Page(s)
LINE COUNT: 1432
CAS INDEXING IS AVAILABLE FOR THIS PATENT.

AB The role of the .epsilon. isozyme of protein kinase C ("**PKC.epsilon.**") in pain perception, particularly hyperalgesia, methods of lessening pain through administration of inhibitors of **PKC.epsilon.**, methods of identifying compounds that modulate pain, and pharmaceutical compositions comprising an inhibitor of **PKC.epsilon.** and **PKC.epsilon** .-independent analgesic agent are disclosed.

CAS INDEXING IS AVAILABLE FOR THIS PATENT.

L19 ANSWER 2 OF 18 EMBASE COPYRIGHT 2002 ELSEVIER SCI. B.V. DUPLICATE 2

ACCESSION NUMBER: 2002146695 EMBASE
TITLE: Inhibition of calcineurin and sarcolemmal Ca(2+) influx protects cardiac morphology and ventricular function in K(v)4.2N transgenic mice.
AUTHOR: Sah R.; Oudit G.Y.; Nguyen T.-T.T.; Lim H.W.; Wickenden A.D.; Wilson G.J.; Molkentin J.D.; Backx P.H.
CORPORATE SOURCE: Dr. P.H. Backx, Heart and Stroke, Richard Lewar Centre, Fitzgerald Building, 150 College St, Toronto, Ont. M55 3EZ, Canada. p.backx@utoronto.ca
SOURCE: Circulation, (16 Apr 2002) 105/15 (1850-1856).
Refs: 35
ISSN: 0009-7322 CODEN: CIRCAZ
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 005 General Pathology and Pathological Anatomy
018 Cardiovascular Diseases and Cardiovascular Surgery
029 Clinical Biochemistry
037 Drug Literature Index
LANGUAGE: English
SUMMARY LANGUAGE: English

AB Background - Cardiac-targeted expression of truncated K(v)4.2 subunit (K(v)4.2N) reduces transient outward current (I(to)) density, prolongs action potentials (APs), and enhances contractility in 3- to 4-week-old **transgenic mice.** By 13 to 15 weeks of age, these mice develop severely impaired cardiac function and signs of heart failure. In this study, we examined whether augmented contractility in K(v)4.2N mice results from elevations in intracellular calcium ([Ca(2+)](i)) secondary to AP prolongation and investigated the putative roles of calcineurin

activation in heart disease development of K(v)4.2N mice. Methods and Results - At 3 to 4 weeks of age, L-type Ca(2+) influx and peak [Ca(2+)](i) were significantly elevated in K(v)4.2N myocytes compared with control because of AP prolongation. Cardiac calcineurin activity was also significantly elevated in K(v)4.2N mice by 5 weeks of age relative to controls and increased progressively as heart disease developed. This was associated with activation of protein kinase C (PKC)-.alpha. and PKC-.theta. but not **PKC-.epsilon.**, as well as increases in .beta.-myosin heavy chain (.beta.-MHC) and reductions in sarcoplasmic/endoplasmic reticulum Ca(2+)-ATPase (SERCA)-2a expression. Treatment with either cyclosporin A or verapamil prevented increases in heart weight to body weight ratios, interstitial fibrosis, impaired contractility, PKC activation, and changes in the expression patterns of .beta.-MHC and SERCA2a. Conclusions - Our results demonstrate that AP prolongation caused by I(lo) reduction results in enhanced Ca(2+) cycling and hypercontractility in mice and suggests that elevations in [Ca(2+)](i) via I(Ca,L) and activation of calcineurin play a central role in disease development after I(lo) reduction using the K(v)4.2N construct.

L19 ANSWER 3 OF 18 CAPLUS COPYRIGHT 2002 ACS DUPLICATE 3

ACCESSION NUMBER: 2001:629761 CAPLUS
DOCUMENT NUMBER: 135:329841
TITLE: Overexpression of manganese superoxide dismutase suppresses tumor formation by modulation of activator protein-1 signaling in a multistage skin carcinogenesis model
AUTHOR(S): Zhao, Yunfeng; Xue, Yi; Oberley, Terry D.; Kiningham, Kelley K.; Lin, Shu-Mei; Yen, Hsiu-Chuan; Majima, Hideyuki; Hines, Judy; St. Clair, Daret
CORPORATE SOURCE: Graduate Center for Toxicology, University of Kentucky, Lexington, KY, 40536, USA
SOURCE: Cancer Research (2001), 61(16), 6082-6088
CODEN: CNREA8; ISSN: 0008-5472
PUBLISHER: American Association for Cancer Research
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Manganese superoxide dismutase (MnSOD) is a nuclear encoded primary antioxidant enzyme localized in mitochondria. Because expression of MnSOD plays a major role in maintaining cellular redox status and reactive oxygen species are known to play a role in signal transduction and carcinogenesis, we investigated the role of MnSOD in the development of cancer using a two-stage [7,12-dimethylbenz(a)-anthracene plus 12-O-tetradecanoylphorbol-13-acetate (TPA)] skin carcinogenesis model. Female transgenic mice expressing the human MnSOD gene in the skin and their nontransgenic counterparts were used in this study. Pathol. examn. demonstrated significant redn. of papilloma formation in transgenic mice. Quant. anal. of 4-hydroxy-2-nonenal-modified proteins showed greater accumulation of oxidative damage products in nontransgenic compared with transgenic mice, and this oxidative damage was demonstrated to be present in both mitochondria and nucleus. TPA increased activator protein-1 (AP-1) binding activity within 6 h in nontransgenic mice, but increased AP-1 binding activity was delayed in the transgenic mice. Electrophoretic mobility shift assay, transcription of the target genes, and Western anal. studies indicated that the increased AP-1 binding activity was attributable to induction of the Jun but not the Fos protein families. Overexpression of MnSOD selectively inhibited the TPA-induced activation of protein kinase C.epsilon. and prevented subsequent activation of c-Jun NH2-terminal kinase in response to TPA. Overall, these results indicate that MnSOD regulates both cellular redox status and selectively modulates **PKC.epsilon.** signaling, thereby delaying AP-1 activation and inhibiting tumor promotion, resulting in redn. of tumors in MnSOD **transgenic mice.**

REFERENCE COUNT: 44 THERE ARE 44 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L19 ANSWER 4 OF 18 CAPLUS COPYRIGHT 2002 ACS DUPLICATE 4

T7-PKC.epsilon. further reduced papilloma burden to 93% compared to wild-type controls but still resulted in the development of squamous-cell carcinoma. To find potential mechanisms of PKC-assocd. differences in tumor promotion, the induction of known downstream effectors of tumor promotion, ornithine decarboxylase (ODC) activity and epidermal hyperplasia, was detd. Despite long-term papilloma inhibition in both PKC.delta. and **PKC.epsilon. transgenic mice**, the induction of ODC by TPA was not attenuated in PKC .delta. and .epsilon. mouse lines. Both PKC transgenic and wild-type mice exhibited sustained hyperplasia after repeated TPA treatments. However, TPA-induced epidermal hyperplasia in T7-PKC.epsilon. mice was significantly increased (52%) compared with T7-PKC.alpha., T7-PKC.delta. and wild-type mice. TPA-induced ODC activity and the resultant accumulation of polyamines may play different roles (e.g., induction of apoptosis vs. proliferation) in the pathways leading to the induction of cancer in PKC.alpha., PKC.delta. and **PKC.epsilon. transgenic mice.**

REFERENCE COUNT: 58 THERE ARE 58 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L19 ANSWER 7 OF 18 EMBASE COPYRIGHT 2002 ELSEVIER SCI. B.V.DUPLICATE 7

ACCESSION NUMBER: 2001060685 EMBASE

TITLE: Increased neuronal and glial expression of protein kinase C isoforms in neocortex of transgenic Tg2576 mice with amyloid pathology.

AUTHOR: Rossner S.; Mehlhorn G.; Schliebs R.; Bigl V.

CORPORATE SOURCE: Dr. S. Rossner, Department of Neurochemistry, Paul Flechsig Inst. for Brain Res., Jahnallee 59, 04109 Leipzig, Germany. rossn@medizin.uni-leipzig.de

SOURCE: European Journal of Neuroscience, (2001) 13/2 (269-278). Refs: 52
ISSN: 0953-816X CODEN: EJONEI

COUNTRY: United Kingdom

DOCUMENT TYPE: Journal; Article

FILE SEGMENT: 008 Neurology and Neurosurgery

LANGUAGE: English

SUMMARY LANGUAGE: English

AB We investigated the influence of five-to sevenfold neuronal overexpression of the Swedish mutation of human APP695 (APPsw) in the **transgenic mouse** strain Tg2576 on neocortical protein kinase C (PKC) expression and subcellular distribution. Using specific antibodies to PKC.alpha., PKC.beta., PKC.gamma., **PKC.epsilon.** and PKC.zeta. isoforms for Western blot analysis, we observed increased immunoreactivity for PKC.alpha. and PKC.gamma. isoforms in crude tissue homogenates from the neocortex of 16-month-old APPsw mice as compared with nontransgenic littermates, which was not present in 6 month-old Tg2576 mice. We also observed elevated levels of PKC.alpha., PKC.beta., PKC.gamma. and PKC.zeta., in membrane fractions and reduced concentrations of PKC.alpha. and PKC.gamma. in cytosolic fractions of aged Tg2576 mice, indicating that these PKC isoforms are in their activated state. In young, 6-month-old Tg2576 mice, however, the increase in membrane-bound PKC isoforms and concomitant decrease in cytosolic PKC isoforms was much less pronounced, demonstrating the age-dependent nature of alterations in PKC isoforms. Immunocytochemistry of brain sections supported these findings and revealed increased neuronal labelling for PKC.alpha., PKC.gamma. and PKC.lambda., isoforms in neocortex of 16-month-old APPsw mice compared with nontransgenic littermates, with the increase being strongest for PKC.gamma. and PKC.lambda. isoforms. Additionally, PKC.gamma. and to a lesser extent PKC.lambda. isoforms were induced in reactive astrocytes in proximity to amyloid plaques. Our data indicate that neuronal overexpression of APPsw causes a dynamic change in neuronal expression and activation of multiple PKC isoforms known to be regulators of proteolytic amyloid precursor protein (APP) processing (PKC.alpha.) and of neuronal survival (PKC.lambda. and PKC.zeta.). The induction of the PKC.gamma. and PKC.lambda. isoforms in reactive astrocytes surrounding amyloid plaques might be required for astrocyte activation and astrocytic cytokine

expression in response to amyloid plaque formation.

L19 ANSWER 8 OF 18 EMBASE COPYRIGHT 2002 ELSEVIER SCI. B.V.
ACCESSION NUMBER: 2001212735 EMBASE
TITLE: RGS4 reduces contractile dysfunction and hypertrophic gene induction in G(.alpha.q) overexpressing mice.
AUTHOR: Rogers J.H.; Tsirka A.; Kovacs A.; Blumer K.J.; Dorn II G.W.; Muslin A.J.
CORPORATE SOURCE: A.J. Muslin, Center for Cardiovascular Research, Box 8086, Washington Univ. School of Medicine, 660 S. Euclid Avenue, St. Louis, MO 63110, United States.
amuslin@imgate.wustl.edu
SOURCE: Journal of Molecular and Cellular Cardiology, (2001) 33/2 (209-218).
Refs: 41
ISSN: 0022-2828 CODEN: JMCDAY
COUNTRY: United Kingdom
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 018 Cardiovascular Diseases and Cardiovascular Surgery
022 Human Genetics
LANGUAGE: English
SUMMARY LANGUAGE: English
AB The intrinsic GTPase activity of G(.alpha.q) is low, and RGS proteins which activate GTPase are expressed in the heart: however, their functional relevance in vivo is unknown. **Transgenic mice** with cardiac-specific overexpression of G(.alpha.q) in myocardium exhibit cardiac hypertrophy, enhanced **PKC.epsilon.** membrane translocation, embryonic gene expression, and depressed cardiac contractility. We recently reported that **transgenic mice** with cardiac-specific expression of RGS4, a G(.alpha.q) and G(.alpha.i) GTPase activator, exhibit decreased left ventricular hypertrophy and ANF induction in response to pressure overload. To test the hypothesis that RGS4 can act as a G(.alpha.q)-specific GTPase activating protein (GAP) in the in vivo heart, dual transgenic G(.alpha.q)-40xRGS4 mice were generated to determine if RGS4 co-expression would ameliorate the G(.alpha.q)-40 phenotype. At age 4 weeks, percent fractional shortening was normalized in dual **transgenic mice** as was left ventricular internal dimension and posterior and septal wall thicknesses. **PKC. epsilon.** membrane translocation and ANF and .alpha.-skeletal actin mRNA levels were also normalized. Compound **transgenic mice** eventually developed depressed cardiac contractility that was evident by 9 weeks of age. These studies establish for the first time a role for RGS4 as a GAP for G(.alpha.q) in the in vivo heart, and demonstrate that its regulated expression can have pathophysiologic consequences.

L19 ANSWER 9 OF 18 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.
ACCESSION NUMBER: 2001:257581 BIOSIS
DOCUMENT NUMBER: PREV200100257581
TITLE: Transgenic activation of PKCepsilon preferentially increases mitochondrial PKCepsilon expression and enhances formation of a mitochondrial PKCepsilon-JNK signaling complex in the mouse heart.
AUTHOR(S): Baines, Christopher P. (1); Zhang, Jun (1); Zheng, Yu-Ting (1); Xiu, Joanne X. (1); Bolli, Roberto (1); Ping, Peipei (1)
CORPORATE SOURCE: (1) University of Louisville, 570 S. Preston St., Baxter Building Suite 122, Louisville, KY, 40202 USA
SOURCE: FASEB Journal, (March 7, 2001) Vol. 15, No. 4, pp. A103. print.
Meeting Info.: Annual Meeting of the Federation of American Societies for Experimental Biology on Experimental Biology 2001 Orlando, Florida, USA March 31-April 04, 2001
ISSN: 0892-6638.
DOCUMENT TYPE: Conference
LANGUAGE: English

SUMMARY LANGUAGE: English

AB Protein kinase C (**PKC**) **epsilon** and the c-Jun N-terminal kinases (JNK) both play crucial roles in the mechanism of ischemic preconditioning (PC). Furthermore, cardiac-specific expression of active PKCepsilon confers a chronically cardioprotected phenotype. Whilst evidence has also implicated mitochondrial KATP channels as end-effectors of PC, how the signal initiated by PKCepsilon is integrated and transmitted to the mitochondria has not been examined. Accordingly, the present study tested the hypothesis that PKCepsilon-induced cardioprotection in **transgenic mice** is associated with preferential upregulation of mitochondrial PKCepsilon and its interaction with JNK. Hearts from either wild-type (NTG) or transgenic (TG) mice with cardiac-specific overexpression of active PKCepsilon (A159E) were homogenized and mitochondrial and cytosolic fractions isolated. The purity of the mitochondrial fraction was verified by immunoblotting with prohibitin-1. Total PKCepsilon expression was increased 6.2 +- 0.3-fold ($P<0.05$) in TG compared to NTG mice. However, this elevation was not equally distributed among the subcellular compartments. Whilst cytosolic levels of PKCepsilon were only moderately elevated 6.2 +- 0.4-fold ($P<0.05$), PKCepsilon was drastically upregulated 34.5 +- 0.9-fold ($P<0.05$) in mitochondria from TG mice. Immunoblotting demonstrated that both JNK1 and JNK2 are present in mitochondria from both NTG and TG mouse hearts with only minimal changes in expression observed in PKCepsilon TG cardiac mitochondria. However, interaction of PKCepsilon with JNK was observed in both NTG and TG mitochondria. Furthermore, upregulation of mitochondrial PKCepsilon increased the amount of PKCepsilon co-precipitating with JNK (45.1 +- 2.9-fold; $P<0.05$) in the mitochondria of 3 out of 5 TG hearts. Taken together, these data show that cardioprotection by transgenic expression of active PKCepsilon is associated with substantially elevated mitochondrial PKCepsilon expression and the upregulation of PKCepsilon-JNK interaction at this subcellular compartment.

L19 ANSWER 10 OF 18 CAPLUS COPYRIGHT 2002 ACS DUPLICATE 8

ACCESSION NUMBER: 2000:34961 CAPLUS
DOCUMENT NUMBER: 132:73661
TITLE: Cells and animals deficient in the .epsilon. isoenzyme of protein kinase C and their use in screening for anxiolytics
INVENTOR(S): Messing, Robert O.; Hodge, Clyde W.
PATENT ASSIGNEE(S): USA
SOURCE: PCT Int. Appl., 98 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2000001805	A1	20000113	WO 1999-US15152	19990702
W: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, SD, SL, SZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG				
AU 9949689	A1	20000124	AU 1999-49689	19990702
EP 1095136	A1	20010502	EP 1999-933688	19990702
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO				

PRIORITY APPLN. INFO.:
US 1998-91755P P 19980706
US 1999-125995P P 19990324
US 1999-340283 A 19990625

US 1998-91755 P 19980706
US 1999-125995 P 19990324
WO 1999-US15152 W 19990702

AB Cells and animals deficient in protein kinase C .epsilon. isoenzyme (**PKC.epsilon.**) that can be used to screen for anti-**anxiety** drugs are described. According to the present invention, **PKC.epsilon.**-inhibiting compds. act in synergy with drugs acting at the GABAA receptor. These modulators of **PKC. epsilon.** may also be used to modulate alc. consumption, self-administration of other drugs of abuse, and the effects of alc. consumption. **PKC.epsilon.** inhibitors may also also be used either alone or in conjunction with allosteric agonists of GABAA receptors, to treat conditions, such as addiction, withdrawal syndrome, skeletal muscle spasms, convulsive seizures, and epilepsy, that are amenable to treatment by allosteric agonists of GABAA receptors. Addnl. aspects of the present invention are diagnostic methods for identifying individuals at risk for becoming alcoholics or abusers of other drugs and kits for performing such diagnostic methods. Transgenic homozygous **PKC.epsilon.** knockout mice were found to show lower levels of **anxiety** than control animals. Gross anatomy of the knockout mice is essentially normal, but there are changes in the patterns of fiber outgrowth and branching in the stratum radiatum. The knockout mice showed lower levels of alc. consumption in ethanol preference drinking tests with a 75% lowering of ethanol preference but did no show any altered preference for sweet (saccharin) or bitter (quinine) flavors or change in general caloric intake. These mice were also hypersensitive to the sedating effects of alc. and to the allosteric GABAA agonists pentobarbital and diazepam.

REFERENCE COUNT: 14 THERE ARE 14 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L19 ANSWER 11 OF 18 EMBASE COPYRIGHT 2002 ELSEVIER SCI. B.V.

ACCESSION NUMBER: 2000169590 EMBASE

TITLE: Calcineurin promotes protein kinase C and c-Jun NH2-terminal kinase activation in the heart. Cross-talk between cardiac hypertrophic signaling pathways.

AUTHOR: De Windt L.J.; Lim H.W.; Haq S.; Force T.; Molkentin J.D.

CORPORATE SOURCE: J.D. Molkentin, Div. of Molec. Cardiovasc. Biology, Dept. of Pediatrics, Children's Hospital Medical Center, 3333 Burnet Ave., Cincinnati, OH 45229-3039, United States. molkj0@chmcc.org

SOURCE: Journal of Biological Chemistry, (5 May 2000) 275/18 (13571-13579).
Refs: 58
ISSN: 0021-9258 CODEN: JBCHA3

COUNTRY: United States

DOCUMENT TYPE: Journal; Article

FILE SEGMENT: 002 Physiology
029 Clinical Biochemistry

LANGUAGE: English

SUMMARY LANGUAGE: English

AB Multiple intracellular signaling pathways have been shown to regulate the hypertrophic growth of cardiomyocytes. Both necessary and sufficient roles have been described for the mitogen activated protein kinase (MAPK) signaling pathway, specific protein kinase C (PKC) isoforms, and calcineurin. Here we investigate the interdependence between calcineurin, MAPK, and PKC isoforms in regulating cardiomyocyte hypertrophy using three separate approaches. Hearts from hypertrophic calcineurin **transgenic mice** were characterized for PKC and MAPK activation. Transgenic hearts demonstrated activation of c-Jun NH2-terminal kinase (JNK) and extracellular signal- regulated kinase (ERK1/2), but not p38 MAPK factors. Calcineurin transgenic hearts demonstrated increased activation of PKC.alpha., .beta., and .theta., but not of .epsilon., .beta.2, or .lambda.. In a second approach, cultured cardiomyocytes were infected with a calcineurin adenovirus to induce hypertrophy and the effects of pharmacologic inhibitors or co-infection

with a dominant negative adenovirus were examined. Calcineurin-mediated hypertrophy was prevented with PKC inhibitors, Ca2+ chelation, and attenuated with a dominant negative SEK-1 (MKK4) adenovirus, but inhibitors of ERK or p38 activation had no effect. In a third approach, we examined the activation of MAPK factors and PKC isoforms during the progression of load-induced hypertrophy in aortic banded rats with or without cyclosporine. We determined that inhibition of calcineurin activity with cyclosporine prevented PKC.alpha. .theta., and JNK activation, but did not affect **PKC.epsilon.**, .beta., .lambda., ERK1/2, or p38 activation. Collectively, these data indicate that calcineurin hypertrophic signaling is interconnected with PKC.alpha., .theta., and JNK in the heart, while **PKC.epsilon.**, .beta., .lambda., p38, and ERK1/2 are not involved in calcineurin-mediated hypertrophy.

L19 ANSWER 12 OF 18 EMBASE COPYRIGHT 2002 ELSEVIER SCI. B.V.DUPLICATE 9
ACCESSION NUMBER: 2001016564 EMBASE
TITLE: The lack of NF-.kappa.B transactivation and PKC.epsilon. expression in CD4(+)CD(+) thymocytes correlates with negative selection.
AUTHOR: Simon A.K.; Auphan N.; Pophillat M.; Boyer C.; Ghosh S.; Rincon M.; Flavell R.A.; Schmitt-Verhulst A.-M.
CORPORATE SOURCE: A.K. Simon, Immunology Group, Institute of Molecular Medicine, John Radcliffe Hospital, Headington, Oxford OX3 9DS, United Kingdom. katja.simon@ndm.ox.ac.uk
SOURCE: Cell Death and Differentiation, (2000) 7/12 (1253-1262). Refs: 64
ISSN: 1350-9047 CODEN: CDDIEK
COUNTRY: United Kingdom
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 026 Immunology, Serology and Transplantation
029 Clinical Biochemistry
LANGUAGE: English
SUMMARY LANGUAGE: English
AB Deletion of autoreactive thymocytes at the DP stage is the basis for tolerance to thymus-expressed self antigens. In this study we investigated whether distinct signalling pathways are induced in DP thymocytes as compared to mature T cells upon stimulation with antigen. Using triple **transgenic mice** expressing a TCR transgene, dominant negative ras/Mek proteins and a reporter gene construct with AP-1 or NF-.kappa.B binding sites, we showed a complete lack of transcriptional activity of NF-.kappa.B but not AP-1 in DP thymocytes, whereas both were transcriptionally active in mature T cells after antigenic stimulation. Lack of NF-.kappa.B induction correlated with increased death in response to antigen. AP-1 induction was dependent on the integrity of the ras/Mek pathway indicating that this pathway was activated in the DP thymocytes. In contrast, we found a complete lack of constitutive expression of the .epsilon. isoform of Protein Kinase C (PKC) in DP thymocytes, although it was present in mature thymocytes and peripheral T cells. Taken together the results suggest that the lack of **PKC.epsilon.** in DP thymocytes could lead to the absence of NF-.kappa.B activity after antigenic stimulation contributing to negative selection.

L19 ANSWER 13 OF 18 CAPLUS COPYRIGHT 2002 ACS DUPLICATE 10
ACCESSION NUMBER: 2000:466544 CAPLUS
DOCUMENT NUMBER: 133:175552
TITLE: Transgenic overexpression of constitutively active protein kinase C .epsilon. causes concentric cardiac hypertrophy
AUTHOR(S): Takeishi, Yasuchika; Ping, Peipei; Bolli, Roberto; Kirkpatrick, Darryl L.; Hoit, Brian D.; Walsh, Richard A.
CORPORATE SOURCE: Department of Medicine, Case Western Reserve University and University Hospitals of Cleveland, Cleveland, OH, 44106-5029, USA
SOURCE: Circulation Research (2000), 86(12), 1218-1223

CODEN: CIRUAL; ISSN: 0009-7330
PUBLISHER: Lippincott Williams & Wilkins
DOCUMENT TYPE: Journal
LANGUAGE: English
AB To test the hypothesis that activation of the protein kinase C (**PKC**) **.epsilon.** isoform leads to cardiac hypertrophy without failure, we studied **transgenic mice** with cardiac-specific overexpression of a constitutively active mutant of the **PKC.epsilon.** isoform driven by an .alpha.-myosin heavy chain promoter. In **transgenic mice**, the protein level of **PKC.epsilon.** in heart tissue was increased 9-fold. There was a 6-fold increase of the membrane/cytosol ratio, and PKC activity in the membrane fraction was 4.2-fold compared with wild-type mice. The heart wt. was increased by 28%, and upregulation of the mRNA for .beta.-myosin heavy chain and .alpha.-skeletal actin was obsd. in transgenic mouse hearts. Echocardiog. demonstrated increased anterior and posterior wall thickness with normal left ventricular function and dimensions, indicating concentric cardiac hypertrophy. Isolated cardiomyocyte mech. function was slightly decreased, and Ca2+ signals were markedly depressed in transgenic mice, suggesting that myofilament sensitivity to Ca2+ was increased. No differences were obsd. in either the levels of cardiac Ca2+-handling proteins or the degree of cardiac regulatory protein phosphorylation between wild-type and transgenic mice. Unlike mice with PKC.beta.2 overexpression, **transgenic mice** with cardiac-specific overexpression of the active **PKC.epsilon.** mutant demonstrated concentric hypertrophy with normal in vivo cardiac function. Thus, PKC isoforms may play differential functional roles in cardiac hypertrophy and failure.
REFERENCE COUNT: 30 THERE ARE 30 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L19 ANSWER 14 OF 18 CAPLUS COPYRIGHT 2002 ACS DUPLICATE 11
ACCESSION NUMBER: 2000:125562 CAPLUS
DOCUMENT NUMBER: 132:263342
TITLE: Transgenic mice overexpressing protein kinase C.epsilon. in their epidermis exhibit reduced papilloma burden but enhanced carcinoma formation after tumor promotion
AUTHOR(S): Reddig, Peter J.; Dreckschmidt, Nancy E.; Zou, Jun; Bourguignon, Sarah E.; Oberley, Terry D.; Verma, Ajit K.
CORPORATE SOURCE: Department of Human Oncology, Medical School, University of Wisconsin, Madison, WI, 53792, USA
SOURCE: Cancer Research (2000), 60(3), 595-602
CODEN: CNREA8; ISSN: 0008-5472
PUBLISHER: AACR Subscription Office
DOCUMENT TYPE: Journal
LANGUAGE: English
AB To det. the role that protein kinase C.epsilon. (**PKC.epsilon.**) may play in skin growth, differentiation, and tumor promotion, **transgenic mice** were generated that overexpressed an epitope-tagged protein kinase C.epsilon. (T7-**PKC.epsilon.**) in their epidermis using the human keratin 14 promoter. Three independent mouse lines that overexpressed the T7-PKC.epsilon. in their epidermis were produced. The three independent lines 206, 224, and 215 exhibited a 3-, 6-, and 18-fold elevation, resp., in the level of PKC.epsilon. immunoreactive protein. Line 215 exhibited a 19-fold greater phosphatidylserine and 12-O-tetradecanoylphorbol-13-acetate (TPA) stimulated kinase activity than line 224. Line 206 exhibited a low basal T7-PKC.epsilon. activity, which failed to be stimulated by phosphatidylserine and TPA. All of the line 215 transgenic mice (F0 to the F2 generation) displayed phenotypic changes in the skin. The phenotypic changes progressed gradually, starting around 4-5 mo of age, with mild dryness of the tail accompanied by hair loss and inflammation at the base of the tail. Hyperproliferation and ulceration of the affected regions were obsd. around 7-8 mo of age. The

hyperproliferative epidermis from the affected regions exhibited an expansion of the suprabasal epidermal cells. Inflammation and/or ulceration were also obsd. in the dorsal skin, the ears, and around the eyes. The line 215 mice, which expressed the highest level of PKC.epsilon., were evaluated for sensitivity to mouse skin tumor promotion by TPA. Tumors were elicited by the initiation (7,12-dimethylbenz[a]anthracene, 100 nmol)-promotion (TPA, 5 nmol/twice weekly) protocol. The papilloma burden was reduced by 95-96% for male and female T7-PKC.epsilon. mice compared to wild-type controls. However, carcinomas developed rapidly in the T7-PKC.epsilon. mice treated with 7, 12-dimethylbenz[a]anthracene and TPA. These carcinomas appeared to form independently of prior papilloma development. These results demonstrate that PKC.epsilon. is an important regulator of skin tumor development.

REFERENCE COUNT: 47 THERE ARE 47 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L19 ANSWER 15 OF 18 CAPLUS COPYRIGHT 2002 ACS DUPLICATE 12

ACCESSION NUMBER: 1999:486847 CAPLUS
DOCUMENT NUMBER: 131:241333
TITLE: Cardiac specific overexpression of transglutaminase II (Gh) results in a unique hypertrophy phenotype independent of phospholipase C activation
AUTHOR(S): Small, Kersten; Feng, Jian-Fang; Lorenz, John; Donnelly, Elizabeth T.; Yu, Andrew; Im, Mie-Jae; Dorn, Gerald W., II; Liggett, Stephen B.
CORPORATE SOURCE: Department of Medicine, University of Cincinnati College of Medicine, Cincinnati, OH, 45267, USA
SOURCE: Journal of Biological Chemistry (1999), 274(30), 21291-21296
CODEN: JBCHA3; ISSN: 0021-9258
PUBLISHER: American Society for Biochemistry and Molecular Biology
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Tissue type transglutaminase (TGII, also known as Gh) has been considered a multifunctional protein, with both transglutaminase and GTPase activity. The role of the latter function, which is proposed as a coupling mechanism between .alpha.1-adrenergic receptors and phospholipase C (PLC), is not well defined. TGII was overexpressed in transgenic mice in a cardiac specific manner to delineated relevant signaling pathways and their consequences in the heart. Cardiac transglutaminase activity in the highest expressing line was .apprx.37-fold greater than in nontransgenic lines. However, in vivo signaling to PLC, as assessed by inositol phosphate turnover in [3H]myoinositol organ bath atrial prepns., was not increased in the TGII mice at base line or in response to .alpha.1-adrenergic receptor stimulation; nor was protein kinase C.alpha. (PKC.alpha.) or **PKC.epsilon.** activity enhanced in the TGII **transgenic mice**. This is in contrast to mice moderately (.apprx.5-fold) overexpressing G.alpha.q, where inositol phosphate turnover and PKC activity were found to be clearly enhanced. TGII overexpression resulted in a remodeling of the heart with mild hypertrophy, elevated expression of .beta.-myosin heavy chain and .alpha.-skeletal actin genes, and diffuse interstitial fibrosis. Resting ventricular function was depressed, but responsiveness to .beta.-agonist was not impaired. This set of pathophysiol. findings is distinct from that evoked by overexpression of G.alpha.q. We conclude that TGII acts in the heart primarily as a transglutaminase, and modulation of this function results in unique pathol. sequelae. Evidence for TGII acting as a G-protein-like transducer of receptor signaling to PLC in the heart is not supported by these studies.

REFERENCE COUNT: 48 THERE ARE 48 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L19 ANSWER 16 OF 18 EMBASE COPYRIGHT 2002 ELSEVIER SCI. B.V.

ACCESSION NUMBER: 1999428321 EMBASE
TITLE: Signal transduction in atria and ventricles of mice with

transient cardiac expression of activated G protein .alpha.(q).

AUTHOR: Mende U.; Kagen A.; Meister M.; Neer E.J.

CORPORATE SOURCE: Dr. U. Mende, Cardiovascular Division, Brigham and Women's Hospital, 75 Francis St, Boston, MA 02115, United States. mende@calvin.bwh.harvard.edu

SOURCE: Circulation Research, (26 Nov 1999) 85/11 (1085-1091). Refs: 32
ISSN: 0009-7330 CODEN: CIRUAL

COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 018 Cardiovascular Diseases and Cardiovascular Surgery
LANGUAGE: English
SUMMARY LANGUAGE: English

AB We recently showed that the transient expression of a hemagglutinin (HA) epitope-tagged, constitutively active mutant of the G protein .alpha.(q) subunit (HA.alpha.(q)*) in the hearts of **transgenic mice** is sufficient to induce cardiac hypertrophy and dilatation that continue to progress after HA.alpha.(q)* protein becomes undetectable. We demonstrated that the activity of phospholipase C.beta., the immediate downstream target of activated G.alpha.(q), is increased at 2 weeks, when HA.alpha.(q)* is expressed, but also at 10 weeks, when HA.alpha.(q)* is no longer detectable. This observation suggested that the transient HA.alpha.(q)* expression causes multiple, persistent changes in cellular signaling pathways. We now demonstrate changes in the level, activity, or both of several signaling components, including changes in the amount and hormone responsiveness of phospholipase C.beta. enzymes, in the basal level of diacylglycerol (which predominantly reflects activation of phospholipase D), in the amount or distribution of protein kinase C (PKC) isoforms (pKC.alpha., PKC.delta., and **PKC.epsilon.**), and in the amount of several endogenous G proteins. These changes vary depending on the isoform of the signaling molecule, the chamber in which it is expressed, and the presence or absence of HA.alpha.(q)*. Our results suggest that a network of linked signaling functions determines the development of hypertrophy. They also suggest that atria and ventricles represent different signaling domains. It is likely that such changes occur in other model systems in which the activity of a single signaling component is increased, either due to an activating mutation or due to overexpression of the wild type.

L19 ANSWER 17 OF 18 CAPLUS COPYRIGHT 2002 ACS DUPLICATE 13

ACCESSION NUMBER: 1999:695607 CAPLUS
DOCUMENT NUMBER: 132:19984
TITLE: Supersensitivity to allosteric GABAA receptor modulators and alcohol in mice lacking **PKC. epsilon.**

AUTHOR(S): Hodge, Clyde W.; Mehmert, Kristin K.; Kelley, Stephen P.; McMahon, Thomas; Haywood, Ashley; Olive, M. Foster; Wang, Dan; Sanchez-Perez, Ana Maria; Messing, Robert O.

CORPORATE SOURCE: Department of Neurology and Ernest Gallo Clinic and Research Center, University of California San Francisco, San Francisco, CA, 94110, USA

SOURCE: Nature Neuroscience (1999), 2(11), 997-1002
CODEN: NANEFN; ISSN: 1097-6256

PUBLISHER: Nature America
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Several of the actions of ethanol are mediated by .gamma.-aminobutyrate type A (GABAA) receptors. Here we demonstrated that mutant mice lacking protein kinase C epsilon (**PKC.epsilon.**) were more sensitive than wild-type littermates to the acute behavioral effects of ethanol and other drugs that allosterically activate GABAA receptors. GABAA receptors in membranes isolated from the frontal cortex of **PKC.epsilon.** null mice were also supersensitive to allosteric activation by ethanol and flunitrazepam. In addn., these

mutant mice showed markedly reduced ethanol self-administration. These findings indicate that inhibition of **PKC.epsilon**. increases sensitivity of GABAA receptors to ethanol and allosteric modulators. Pharmacol. agents that inhibit **PKC.epsilon**. may be useful for treatment of alcoholism and may provide a non-sedating alternative for enhancing GABAA receptor function to treat other disorders such as **anxiety** and epilepsy.

REFERENCE COUNT: 46 THERE ARE 46 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L19 ANSWER 18 OF 18 EMBASE COPYRIGHT 2002 ELSEVIER SCI. B.V.DUPLICATE 14

ACCESSION NUMBER: 96112094 EMBASE
DOCUMENT NUMBER: 1996112094
TITLE: Endothelin-dependent actions in cultured AT-1 cardiac myocytes: The role of the .epsilon. isoform of protein kinase C.
AUTHOR: Jiang T.; Pak E.; Zhang H.; Kline R.P.; Steinberg S.F.
CORPORATE SOURCE: Department of Medicine, College of Physicians and Surgeons, Columbia University, 630 W 168 St,New York, NY 10032, United States
SOURCE: Circulation Research, (1996) 78/4 (724-736).
ISSN: 0009-7330 CODEN: CIRUAL
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 018 Cardiovascular Diseases and Cardiovascular Surgery
029 Clinical Biochemistry
LANGUAGE: English
SUMMARY LANGUAGE: English

AB The consequences of endothelin receptor activation were examined in atrial tumor myocytes derived from **transgenic mice** (AT-1 cells). Endothelin-1 (endothelin) stimulates phosphoinositide hydrolysis in a dose-dependent manner. Endothelin also induces the rapid and transient translocation of protein kinase C (**PKC**)-. **epsilon**. immunoreactivity from the soluble to the particulate cell fraction. The subcellular distributions of PKC.alpha. and PKC.zeta. (also expressed by AT-1 cells) are not influenced by endothelin. Using quantitative fluorescence microscopy with fura 2, we examined the effects of endothelin on intracellular calcium. In electrically driven myocytes, endothelin induces a rapid and transient increase in the amplitude of the calcium transient. This is blocked by both phorbol 12-myristate 13-acetate (PMA) pretreatment to downregulate PKC and the PKC inhibitor chelerythrine, arguing the **PKC.epsilon**. plays a critical role in endothelin receptor-dependent increases in intracellular calcium. Endothelin also stimulates mitogen- activated protein kinase (MAPK). MAPK activation is markedly attenuated by pretreatment with PMA or pertussis toxin (PTX, to inactivate susceptible G protein .alpha. subunits); it is completely prevented by combined pretreatment with PMA and PTX. In contrast, it is not attenuated by chelation of intracellular calcium with BAPTA. These findings indicate that the pathway for endothelin receptor stimulation of MAPK involves **PKC**. **epsilon**. and PTX-sensitive G protein(s). Thus, these studies identify a functional role for **PKC.epsilon**. as a mediator of endothelin receptor-dependent increases in cytosolic calcium and MAPK activity in AT-1 cells. Accordingly, the AT-1 cell system should provide a uniquely useful model to identify the intracellular targets for **PKC.epsilon**. and investigate their function in the regulation of intracellular calcium homeostasis and the induction of the growth response in cardiac myocytes.

=> d 1-6 ibib abs

L14 ANSWER 1 OF 6 MEDLINE
ACCESSION NUMBER: 2002219232 MEDLINE
DOCUMENT NUMBER: 21952562 PubMed ID: 11956130
TITLE: Inhibition of calcineurin and sarcolemmal Ca2+ influx protects cardiac morphology and ventricular function in K(v)4.2N transgenic mice.
AUTHOR: Sah Rajan; Oudit Gavin Y; Nguyen The-Tin T; Lim Hae W; Wickenden Alan D; Wilson Gregory J; Molkentin Jeffery D; Backx Peter H
CORPORATE SOURCE: Department of Physiology and the Division of Cardiology, University Health Network, University of Toronto, Toronto, Canada.
SOURCE: CIRCULATION, (2002 Apr 16) 105 (15) 1850-6.
Journal code: 0147763. ISSN: 1524-4539.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Abridged Index Medicus Journals; Priority Journals
ENTRY MONTH: 200204
ENTRY DATE: Entered STN: 20020417
Last Updated on STN: 20020426
Entered Medline: 20020425

AB BACKGROUND: Cardiac-targeted expression of truncated K(v)4.2 subunit (K(v)4.2N) reduces transient outward current (I(to)) density, prolongs action potentials (APs), and enhances contractility in 3- to 4-week-old **transgenic mice**. By 13 to 15 weeks of age, these mice develop severely impaired cardiac function and signs of heart failure. In this study, we examined whether augmented contractility in K(v)4.2N mice results from elevations in intracellular calcium ([Ca2+]i) secondary to AP prolongation and investigated the putative roles of calcineurin activation in heart disease development of K(v)4.2N mice. METHODS AND RESULTS: At 3 to 4 weeks of age, L-type Ca2+ influx and peak [Ca2+]i were significantly elevated in K(v)4.2N myocytes compared with control because of AP prolongation. Cardiac calcineurin activity was also significantly elevated in K(v)4.2N mice by 5 weeks of age relative to controls and increased progressively as heart disease developed. This was associated with activation of protein kinase C (PKC)-alpha and PKC-theta but not **PKC-epsilon**, as well as increases in beta-myosin heavy chain (beta-MHC) and reductions in sarcoplasmic/endoplasmic reticulum Ca2+-ATPase (SERCA)-2a expression. Treatment with either cyclosporin A or verapamil prevented increases in heart weight to body weight ratios, interstitial fibrosis, impaired contractility, PKC activation, and changes in the expression patterns of beta-MHC and SERCA2a. CONCLUSIONS: Our results demonstrate that AP prolongation caused by I(to) reduction results in enhanced Ca2+ cycling and hypercontractility in mice and suggests that elevations in [Ca2+]i via I(Ca,L) and activation of calcineurin play a central role in disease development after I(to) reduction using the K(v)4.2N construct.

L14 ANSWER 2 OF 6 MEDLINE
ACCESSION NUMBER: 2001429556 MEDLINE
DOCUMENT NUMBER: 21369799 PubMed ID: 11477572
TITLE: Relation of the induction of epidermal ornithine decarboxylase and hyperplasia to the different skin tumor-promotion susceptibilities of protein kinase C alpha, -delta and -epsilon transgenic mice.
AUTHOR: Jansen A P; Dreckschmidt N E; Verwiebe E G; Wheeler D L; Oberley T D; Verma A K
CORPORATE SOURCE: Department of Human Oncology, Medical School, University of Wisconsin, Madison, WI, USA.
CONTRACT NUMBER: RO1 CA35368 (NCI)
SOURCE: INTERNATIONAL JOURNAL OF CANCER, (2001 Sep 1) 93 (5) 635-43.
Journal code: 0042124. ISSN: 0020-7136.

Last Updated on STN: 20010502
Entered Medline: 20010426

AB Deletion of autoreactive thymocytes at the DP stage is the basis for tolerance to thymus-expressed self antigens. In this study we investigated whether distinct signalling pathways are induced in DP thymocytes as compared to mature T cells upon stimulation with antigen. Using triple **transgenic mice** expressing a TCR transgene, dominant negative ras/Mek proteins and a reporter gene construct with AP-1 or NF-kappa B binding sites, we showed a complete lack of transcriptional activity of NF-kappa B but not AP-1 in DP thymocytes, whereas both were transcriptionally active in mature T cells after antigenic stimulation. Lack of NF-kappa B induction correlated with increased death in response to antigen. AP-1 induction was dependent on the integrity of the ras/Mek pathway indicating that this pathway was activated in the DP thymocytes. In contrast, we found a complete lack of constitutive expression of the epsilon isoform of Protein Kinase C (PKC) in DP thymocytes, although it was present in mature thymocytes and peripheral T cells. Taken together the results suggest that the lack of **PKC epsilon** in DP thymocytes could lead to the absence of NF-kappa B activity after antigenic stimulation contributing to negative selection. Cell Death and Differentiation (2000) 7, 1253 - 1262.

L14 ANSWER 4 OF 6 MEDLINE
ACCESSION NUMBER: 2001170646 MEDLINE
DOCUMENT NUMBER: 21097297 PubMed ID: 11168531
TITLE: Increased neuronal and glial expression of protein kinase C isoforms in neocortex of transgenic Tg2576 mice with amyloid pathology.
AUTHOR: Rossner S; Mehlhorn G; Schliebs R; Bigl V
CORPORATE SOURCE: Department of Neurochemistry, Paul Flechsig Institute for Brain Research, Jahnallee 59, 04109 Leipzig, Germany.. rossn@medizin.uni-leipzig.de
SOURCE: EUROPEAN JOURNAL OF NEUROSCIENCE, (2001 Jan) 13 (2) 269-78. Journal code: 8918110. ISSN: 0953-816X.
PUB. COUNTRY: France
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200105
ENTRY DATE: Entered STN: 20010517
Last Updated on STN: 20010517
Entered Medline: 20010510

AB We investigated the influence of five- to sevenfold neuronal overexpression of the Swedish mutation of human APP695 (APPsw) in the **transgenic mouse** strain Tg2576 on neocortical protein kinase C (PKC) expression and subcellular distribution. Using specific antibodies to PKC alpha, PKC beta, PKC gamma, **PKC epsilon** and PKC zeta isoforms for Western blot analysis, we observed increased immunoreactivity for PKC alpha and PKC gamma isoforms in crude tissue homogenates from the neocortex of 16-month-old APPsw mice as compared with nontransgenic littermates, which was not present in 6 month-old Tg2576 mice. We also observed elevated levels of PKC alpha, PKC beta, PKC gamma and PKC zeta in membrane fractions and reduced concentrations of PKC alpha and PKC gamma in cytosolic fractions of aged Tg2576 mice, indicating that these PKC isoforms are in their activated state. In young, 6-month-old Tg2576 mice, however, the increase in membrane-bound PKC isoforms and concomitant decrease in cytosolic PKC isoforms was much less pronounced, demonstrating the age-dependent nature of alterations in PKC isoforms. Immunocytochemistry of brain sections supported these findings and revealed increased neuronal labelling for PKC alpha, PKC gamma and PKC lambda isoforms in neocortex of 16-month-old APPsw mice compared with nontransgenic littermates, with the increase being strongest for PKC gamma and PKC lambda isoforms. Additionally, PKC gamma and to a lesser extent PKC lambda isoforms were induced in reactive astrocytes in proximity to amyloid plaques. Our data indicate that neuronal overexpression of APPsw causes a dynamic change in neuronal

transgenic mice, which overexpress (.apprx.18-fold) epitope-tagged protein kinase C-.epsilon. (T7-**PKC. epsilon.**) protein in the epidermis provide a unique murine model system for highly malignant/metastatic SCC. Skin tumors were developed by the initiation-promotion protocol (initiation with 100 nmol 7,12-dimethyl-benz[a]anthracene; promotion with 5 nmol 12-O-tetradecanoylphorbol-13-acetate twice weekly). T7-**PKC. epsilon. transgenic mice** showed 92% suppression of papilloma development compared with wild-type littermates after 23 wk of tumor promotion. However, within 15-20 wk of 12-O-tetradecanoylphorbol-13-acetate promotion, 40% of T7-PKC.epsilon. mice developed at least one carcinoma compared with 7% of the wild-type mice. All carcinomas from T7-PKC.epsilon. mice appeared without prior papilloma formation. Interestingly, 7,12-dimethyl-benz[a]anthracene alone resulted in the development of squamous cell carcinomas in 22% of T7-PKC.epsilon. mice, whereas wild-type littermates developed no tumors. Histopathol. anal. of tumors from multiple T7-PKC.epsilon. mice revealed moderately differentiated SCC invading the dermal region with neoplasia appearing to originate and invade from the hair follicle. Carcinomas of T7-PKC.epsilon. mice rapidly metastasized to regional lymph nodes within 3 wk of appearance. In wild-type mice, the grade of the invading tumors, originating from interfollicular epidermis, was pathol. categorized as well-differentiated SCC and remained localized to the dermis. The T7-**PKC.epsilon. transgenic mice** may provide a rapid and unique in vivo model to investigate metastatic SCC.

REFERENCE COUNT: 20 THERE ARE 20 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L19 ANSWER 6 OF 18 CAPLUS COPYRIGHT 2002 ACS DUPLICATE 6

ACCESSION NUMBER: 2001:561341 CAPLUS
DOCUMENT NUMBER: 135:270965
TITLE: Relation of the induction of epidermal ornithine decarboxylase and hyperplasia to the different skin tumor-promotion susceptibilities of protein kinase C.alpha., -.delta. and -.epsilon. transgenic mice
AUTHOR(S): Jansen, Aaron P.; Dreckschmidt, Nancy E.; Verwiebe, Eric G.; Wheeler, Deric L.; Oberley, Terry D.; Verma, Ajit K.
CORPORATE SOURCE: Department of Human Oncology, Medical School, University of Wisconsin, Madison, WI, USA
SOURCE: International Journal of Cancer (2001), 93(5), 635-643
CODEN: IJCNAW; ISSN: 0020-7136
PUBLISHER: Wiley-Liss, Inc.
DOCUMENT TYPE: Journal
LANGUAGE: English

AB To define the in vivo role of individual PKC isoforms in mouse skin carcinogenesis, the authors previously characterized FVB/n **transgenic mice** that over-expressed epitope-tagged PKC.delta. (T7-PKC.delta.) or **PKC.epsilon.** (T7-**PKC.epsilon.**) isoforms under the regulation of the human K14 promoter. In continuation of the authors' prior PKC isoform specificity studies, the authors now report the generation of FVB/n transgenic mice with K14-regulated, epitope-tagged PKC.alpha. (T7-PKC.alpha.). T7-PKC.alpha. transgenic mice (line 115) express 8-fold more PKC.alpha. protein than wild-type mice. Using high-resoln. immunogold cytochem., the authors detd. that transgenic over-expression of T7-PKC.alpha. did not alter the subcellular localization of PKC.alpha. but that the d. of PKC.alpha. staining increased. PKC.alpha. localized primarily to the cytoskeleton (tonofilaments, tight junctions) and cell membranes, with modest but definite nuclear labeling also identified. Also, PKC.alpha. over-expression did not alter the immunoreactive protein levels of other PKC isoforms (.delta., .epsilon., .eta., .zeta., .mu.) in the epidermis. Skin tumor-promotion susceptibility was compared among all 3 lines of T7-PKC transgenic mice (.alpha., .delta. and .epsilon.). While T7-PKC.alpha. had no effect on skin tumor promotion by TPA, T7-PKC.delta. reduced papilloma burden by 76% compared to wild-type controls.

ACCESSION NUMBER: 2001:309329 CAPLUS
DOCUMENT NUMBER: 135:17297
TITLE: PKC.epsilon. activation induces dichotomous cardiac phenotypes and modulates PKC.epsilon.-RACK interactions and RACK expression
AUTHOR(S): Pass, Jason M.; Zheng, Yuting; Wead, William B.; Zhang, Jun; Li, Richard C. X.; Bolli, Roberto; Ping, Peipei
CORPORATE SOURCE: Department of Physiology and Biophysics, University of Louisville, Louisville, KY, 40292, USA
SOURCE: American Journal of Physiology (2001), 280(3, Pt. 2), H946-H955
CODEN: AJPHAP; ISSN: 0002-9513
PUBLISHER: American Physiological Society
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Receptors for activated C kinase (RACKs) have been shown to facilitate activation of protein kinase C (PKC). However, it is unknown whether PKC activation modulates RACK protein expression and PKC-RACK interactions. This issue was studied in two PKC.epsilon. transgenic lines exhibiting dichotomous cardiac phenotypes: one exhibits increased resistance to myocardial ischemia (cardioprotected phenotype) induced by a modest increase in PKC.epsilon. activity (288 .+-. 23% of control), whereas the other exhibits cardiac hypertrophy and failure (hypertrophied phenotype) induced by a marked increase in PKC.epsilon. activity (452 .+-. 28% of control). Our data demonstrate that activation of PKC modulates the expression of RACK isotypes and PKC-RACK interactions in a PKC.epsilon. activity- and dosage-dependent fashion. We found that, in mice displaying the cardioprotected phenotype, activation of PKC.epsilon. enhanced RACK2 expression (178 .+-. 13% of control) and particulate PKC.epsilon.-RACK2 protein-protein interactions (178 .+-. 18% of control). In contrast, in mice displaying the hypertrophied phenotype, there was not only an increase in RACK2 expression (330 .+-. 33% of control) and particulate PKC.epsilon.-RACK2 interactions (154 .+-. 14% of control) but also in RACK1 protein expression (174 .+-. 10% of control). Most notably, PKC.epsilon.-RACK1 interactions were identified in this line. With the use of **transgenic mice** expressing a dominant neg. **PKC.epsilon.**, we found that the changes in RACK expression as well as the attending cardiac phenotypes were dependent on **PKC.epsilon.** activity. Our observations demonstrate that RACK expression is dynamically regulated by PKC.epsilon. and suggest that differential patterns of PKC.epsilon.-RACK interactions may be important determinants of PKC.epsilon.-dependent cardiac phenotypes.

REFERENCE COUNT: 57 THERE ARE 57 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L19 ANSWER 5 OF 18 CAPLUS COPYRIGHT 2002 ACS DUPLICATE 5
ACCESSION NUMBER: 2001:150858 CAPLUS
DOCUMENT NUMBER: 134:324385
TITLE: Protein kinase C-.epsilon. transgenic mice: a unique model for metastatic squamous cell carcinoma
AUTHOR(S): Jansen, Aaron P.; Verwiebe, Eric G.; Dreckschmidt, Nancy E.; Wheeler, Deric L.; Oberley, Terry D.; Verma, Ajit K.
CORPORATE SOURCE: Department of Human Oncology, Medical School, University of Wisconsin, Madison, WI, 53792, USA
SOURCE: Cancer Research (2001), 61(3), 808-812
CODEN: CNREA8; ISSN: 0008-5472
PUBLISHER: American Association for Cancer Research
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Squamous cell carcinoma (SCC) and basal cell carcinoma (BCC) are the most common forms of human skin cancer. BCC is slow growing and mostly localized, whereas SCC metastasizes to the regional lymph nodes and subsequently to distal organs. In murine skin carcinogenesis models for SCC, the incidence of metastasis is very low. We report here that FVB/N

expression and activation of multiple PKC isoforms known to be regulators of proteolytic amyloid precursor protein (APP) processing (PKC alpha) and of neuronal survival (PKC lambda and PKC zeta). The induction of the PKC gamma and PKC lambda isoforms in reactive astrocytes surrounding amyloid plaques might be required for astrocyte activation and astrocytic cytokine expression in response to amyloid plaque formation.

L14 ANSWER 5 OF 6 MEDLINE
ACCESSION NUMBER: 2000325059 MEDLINE
DOCUMENT NUMBER: 20325059 PubMed ID: 10864911
TITLE: Transgenic overexpression of constitutively active protein kinase C epsilon causes concentric cardiac hypertrophy.
AUTHOR: Takeishi Y; Ping P; Bolli R; Kirkpatrick D L; Hoit B D; Walsh R A
CORPORATE SOURCE: Department of Medicine, Case Western Reserve University and University Hospitals of Cleveland, Cleveland, Ohio 44106-5029, USA.
CONTRACT NUMBER: HL-43151 (NHLBI)
HL-52318 (NHLBI)
HL-58166 (NHLBI)
+
SOURCE: CIRCULATION RESEARCH, (2000 Jun 23) 86 (12) 1218-23.
Journal code: 0047103. ISSN: 1524-4571.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200008
ENTRY DATE: Entered STN: 20000811
Last Updated on STN: 20010521
Entered Medline: 20000801

AB To test the hypothesis that activation of the protein kinase C (**PKC**) **epsilon** isoform leads to cardiac hypertrophy without failure, we studied **transgenic mice** with cardiac-specific overexpression of a constitutively active mutant of the PKCepsilon isoform driven by an alpha-myosin heavy chain promoter. In **transgenic mice**, the protein level of PKCepsilon in heart tissue was increased 9-fold. There was a 6-fold increase of the membrane/cytosol ratio, and PKC activity in the membrane fraction was 4.2-fold compared with wild-type mice. The heart weight was increased by 28%, and upregulation of the mRNA for beta-myosin heavy chain and alpha-skeletal actin was observed in **transgenic mouse** hearts. Echocardiography demonstrated increased anterior and posterior wall thickness with normal left ventricular function and dimensions, indicating concentric cardiac hypertrophy. Isolated cardiomyocyte mechanical function was slightly decreased, and Ca(2+) signals were markedly depressed in **transgenic mice**, suggesting that myofilament sensitivity to Ca(2+) was increased. No differences were observed in either the levels of cardiac Ca(2+)-handling proteins or the degree of cardiac regulatory protein phosphorylation between wild-type and **transgenic mice**. Unlike mice with PKCbeta(2) overexpression, **transgenic mice** with cardiac-specific overexpression of the active PKCepsilon mutant demonstrated concentric hypertrophy with normal in vivo cardiac function. Thus, PKC isoforms may play differential functional roles in cardiac hypertrophy and failure.

L14 ANSWER 6 OF 6 MEDLINE
ACCESSION NUMBER: 96183785 MEDLINE
DOCUMENT NUMBER: 96183785 PubMed ID: 8635230
TITLE: Endothelin-dependent actions in cultured AT-1 cardiac myocytes. The role of the epsilon isoform of protein kinase C.
AUTHOR: Jiang T; Pak E; Zhang H L; Kline R P; Steinberg S F
CORPORATE SOURCE: Department of Medicine,Columbia University, New York 10032, USA.
CONTRACT NUMBER: 07271 (NHLBI)

PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200108
ENTRY DATE: Entered STN: 20010820
Last Updated on STN: 20010820
Entered Medline: 20010816

AB To define the in vivo role of individual PKC isoforms in mouse skin carcinogenesis, we previously characterized FVB/n **transgenic mice** that over-expressed epitope-tagged PKC delta (T7-PKC delta) or **PKC epsilon** (T7-**PKC epsilon**) isoforms under the regulation of the human K14 promoter. In continuation of our prior PKC isoform specificity studies, we now report the generation of FVB/n **transgenic mice** with K14-regulated, epitope-tagged PKC alpha (T7-PKC alpha). T7-PKC alpha **transgenic mice** (line 115) express 8-fold more PKC alpha protein than wild-type mice. Using high-resolution immunogold cytochemistry, we determined that transgenic over-expression of T7-PKC alpha did not alter the subcellular localization of PKC alpha but that the density of PKC alpha staining increased. PKC alpha localized primarily to the cytoskeleton (tonofilaments, tight junctions) and cell membranes, with modest but definite nuclear labeling also identified. Also, PKC alpha over-expression did not alter the immunoreactive protein levels of other PKC isoforms (delta, epsilon, eta, zeta, mu) in the epidermis. Skin tumor-promotion susceptibility was compared among all 3 lines of T7-PKC **transgenic mice** (alpha, delta and epsilon). While T7-PKC alpha had no effect on skin tumor promotion by TPA, T7-PKC delta reduced papilloma burden by 76% compared to wild-type controls. T7-**PKC epsilon** further reduced papilloma burden to 93% compared to wild-type controls but still resulted in the development of squamous-cell carcinoma. To find potential mechanisms of PKC-associated differences in tumor promotion, the induction of known downstream effectors of tumor promotion, ornithine decarboxylase (ODC) activity and epidermal hyperplasia, was determined. Despite long-term papilloma inhibition in both PKC delta and **PKC epsilon transgenic mice**, the induction of ODC by TPA was not attenuated in PKC delta and epsilon mouse lines. Both PKC transgenic and wild-type mice exhibited sustained hyperplasia after repeated TPA treatments. However, TPA-induced epidermal hyperplasia in T7-**PKC epsilon** mice was significantly increased (52%) compared with T7-PKC alpha, T7-PKC delta and wild-type mice. TPA-induced ODC activity and the resultant accumulation of polyamines may play different roles (e.g., induction of apoptosis vs. proliferation) in the pathways leading to the induction of cancer in PKC alpha, PKC delta and **PKC epsilon transgenic mice**.

L14 ANSWER 3 OF 6 MEDLINE
ACCESSION NUMBER: 2001224669 MEDLINE
DOCUMENT NUMBER: 21110357 PubMed ID: 11175263
TITLE: The lack of NF-kappa B transactivation and PKC epsilon expression in CD4(+)CD8(+) thymocytes correlates with negative selection.
AUTHOR: Simon A K; Auphan N; Pophillat M; Boyer C; Ghosh S; Rincon M; Flavell R A; Schmitt-Verhulst A M
CORPORATE SOURCE: Centre d'Immunologie INSERM-CNRS de Marseille Luminy, Marseille, France.. katja.simon@ndm.ox.ac.uk
SOURCE: CELL DEATH AND DIFFERENTIATION, (2000 Dec) 7 (12) 1253-62. Journal code: 9437445. ISSN: 1350-9047.
PUB. COUNTRY: England: United Kingdom
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200104
ENTRY DATE: Entered STN: 20010502